In the Specification

Please amend the title of the International Patent Application to read as follows:

~~THERAPEUTIC PREPARATION FOR HEMATOPOIETIC DISEASE~~

THERAPEUTIC FORMULATIONS FOR HEMATOPOIETIC DISEASES

Please add the following paragraph at page 1, above line 5 after the Title:

This application is a National Stage Application of International Application Number PCT/JP2004/011951, filed August 13, 2004; which claims priority to U.S. Provisional Application No. 60/495,001.

Please amend the paragraph beginning at page 1, line 10- page 2, line 13 (Background Art) as follows:

Quality control of proteins in endoplasmic reticulum (ER) and transcriptional control of the amount of proteins in the nucleus are important processes that maintain cellular homeostasis (Hampton, R.Y., 2002, "ER-associated degradation in protein quality control and cellular regulation." Curr. Opin. Cell Biol. 14: 476-482). In eukaryotic cells, newly synthesized proteins are transported into the ER where they are correctly folded. However, various environmental conditions, such as large amounts of protein influx into the ER, could trigger a cellular response called unfolded protein response [[(URP)]] (UPR) to overcome this problem (Welihinda, A.A., Tirasophon, W., and Kaufman, R.J., 1999, "The cellular response to protein misfolding in the endoplasmic reticulum." Gene Expr. 7: 293-300). During the [[URP]] UPR response, synthesis of new proteins is globally inhibited by inactivation of eukaryotic initiation factor (eIF) 2α to reduce additional accumulation of misfolded proteins in ER, and genes encoding the ER chaperone proteins including Bip/Grp78 and Grp94, are

also upregulated to re-fold the misfolded proteins correctly (Ron, D., 2002, "Translational control in the endoplasmic reticulum stress response." J. Clin. Invest. 110: 1383-1388). When the amount of misfolded proteins exceeds the protein folding capacity, in spite of [[URP]] UPR, misfolded proteins are eliminated by ubiquitin- and proteasome-dependent degradation processes, known as ER-associated degradation (ERAD) (Hampton, 2002, supra). Misfolded proteins in the ER are translocated into the cytosol, where they become targets of 26S proteasome by ubiquitin ligase enzymes. Various ubiquitin ligases are reported in the ERAD system in mammalian cells, including CHIP (C-terminus of Hsc70-interacting protein) (Ballinger, C.A., Connell, P., Wu, Y., Hu, Z., Thompson, L.J., Yin, L.Y., and Patterson, C., 1999, "Identification of CHIP, a novel tetratricopeptide repeat-containing protein that interacts with heat shock proteins and negatively regulates chaperone functions." Mol. Cell Biol. 19: 4535-4545; Meacham, G.C., Patterson, C., Zhang, W., Younger, J.M., and Cyr, D.M., 2001, "The Hsc70 co-chaperone CHIP targets immature CFTR for proteasomal degradation." Nat. Cell Biol. 3: 100-105; Imai, Y., Soda, M., Inoue, H., Hattori, N., Mizuno, Y., and Takahashi, R., 2001, "An Unfolded Putative Transmembrane Polypeptide, which Can Lead Endoplasmic Reticulum Stress, Is a Substrate of Parkin." Cell 105: 891-902), parkin (Imai, Y., Soda, M., Hatakeyama, S., Akagi, T., Hashikawa, T., Nakayama, K.I., and Takahashi, R., 2002, "CHIP is associated with Parkin, a gene responsible for familial Parkinson's disease, and enhances its ubiquitin ligase activity." Mol. Cell 10: 55-67), gp78/AMFR (Shimizu, K., Tani, M., Watanabe, H., Nagamachi, Y., Niinaka, Y., Shiroishi, T., Ohwada, S., Raz, A., and Yokota, J., 1999, "The autocrine motility factor receptor gene encodes a novel type of seven transmembrane protein." FEBS Lett. 456: 295-300; Fang, S., Ferrone, M., Yang, C., Jensen, J.P., Tiwari, S., and Weissman, A.M., 2001, "The tumor autocrine motility factor receptor, gp78, is a ubiquitin protein ligase implicated in degradation from the endoplasmic reticulum." Proc. Natl. Acad. Sci. U.S.A. 98: 14422-

14427) and Fbx2/FBG1/NFB42 (Yoshida, Y., Chiba, T., Tokunaga, F., Kawasaki, H., Iwai, K., Suzuki, T., Ito, Y., Matsuoka, K., Yoshida, M., Tanaka, K., and Tai, T., 2002, "E3 ubiquitin ligase that recognizes sugar chains." Nature 418: 438-442). Intensive research is currently being conducted to determine the precise mechanisms that regulate the ERAD system in the ER.

Please amend the paragraph at page 3, lines 6-11 as follows:

Recently, by immunoscreening using anti-synovial cell antibodies, the present inventors cloned synoviolin~~/HRD1 (herein after abbreviated as "synoviolin")~~, a human homologue of yeast ubiquitin ligase (E3) ~~Hrd1p/Del3~~ Hrd1p/Del3p (Bays, N.W., Gardner, R.G., Seelig, L.P., Joazeiro, C.A., and Hampton, R.Y., 2001, "Hrd1p/Der3p is a membrane-anchored ubiquitin ligase required for ER-associated degradation." Nat. Cell Biol. 3: 24-29), which is an ER resident membrane protein that comprises a RING-H2 motif.

Please amend the paragraph at page 4, line 28 –33 as follows:

Based on the above-mentioned findings, the present invention provides the following therapeutic pharmaceutical agents and therapeutic methods for hematopoietic diseases, and methods of screening for pharmaceutical agents for treatment of hematopoietic diseases.

[1] A method for treating hematopoietic diseases, wherein the method comprises administering any one of the proteins selected from the group consisting of ~~(a) to (b)~~ (a) to (d) shown below, or a polynucleotide encoding the protein:

Please amend the paragraph beginning at page 14, lines 17- 26 as follows:

In addition to gene targeting as a means for deleting synoviolin function, inhibiting the post-transcription expression without modifying the synoviolin gene on chromosomal DNA, such as the antisense method, ribozyme method, or [[iRNA]] RNAi method may be used as the means to turn off synoviolin function. In the antisense method, a vector comprising a DNA encoding an RNA complementary to the transcription product of a DNA encoding the protein of the present invention is used. In the ribozyme method, for example, a vector comprising a DNA encoding an RNA that cleaves the transcription product of a DNA encoding the protein of the present invention is used. The vector is introduced into mammalian embryonic stem cells as described above, and these cells are injected into mammalian embryos, from which individuals can then be obtained.

Please amend the paragraph beginning at page 14, lines 27- 32 as follows:

In the [[iRNA]] RNAi method, a vector for expressing a DNA encoding the synoviolin protein, for example, a double stranded RNA is introduced into mammalian embryonic stem cells as described above, wherein the antisense RNA complementary to a transcription product comprising the sequence of SEQ ID NO: 1 is paired with a sense RNA complementary to this antisense RNA, and these cells are inserted into mammalian embryos, from which individuals can then be obtained.

Please amend the paragraph beginning at page 16, line 28- page 17, line 11 (Brief Description of the Drawings) as follows:

Fig. 1 is a set of diagrams and photographs showing targeted disruption of the synoviolin gene. Fig. 1(A) shows the structure of the synoviolin wild-type allele, the targeting vector, targeted allele, and partial restriction enzyme map of the genes before and

after targeting events. Exons of the gene are shown as closed boxes, and the β-galactosidase gene (LacZ), neomycin phosphotransferase gene (Neo), diphtheria toxin-A gene (DT), and pBluescript II (BSK) are shown as open boxes. The restriction sites used are indicated as: B for *Bgl*II; P for *Pst*I; E for *EcoR*I; X for *Xho*I; and N for *Nco*I. Fig. 1(B) is a set of photographs showing the Southern blot analysis of targeted ES clones and embryonic clones generated by crossing heterozygous knockout (hereinafter denoted as "syno$^{+/-}$") mice with each other. Genomic DNA from wild-type (hereinafter denoted as "syno$^{+/+}$") TT2 ES cells (WT) and homologously targeted ES clones (clone-1, clone-2) were digested with *Bgl*II and hybridized with an external probe. The syno$^{+/+}$ and syno$^{+/-}$ alleles produced 7.4 kb and 11.7 kb fragments, respectively. Chromosomal DNA isolated from E13.0 embryos generated by crossing syno$^{+/-}$ mice was digested with *Pst*I, and hybridized using the same probe as described above. Fig. 1(C) is a photograph of Northern blot analysis. Twenty μg of total RNA, isolated from E13.0 embryos generated by crossing *syno*$^{+/-}$ mice, was hybridized with a synoviolin probe or glyceraldehyde-3-phosphate dehydrogenase (G3PDH) probe. Fig. 1(D) is a photograph showing the result of isolating proteins from E13.0 embryos and separating them by ~~SES-PAGE~~ SDS-PAGE (50 μg protein/lane). The proteins were transferred to a membrane, and then reacted with an anti-synoviolin antibody.

Please amend the paragraph beginning at page 17, lines 17-19 as follows:

Fig. 3 is a set of photographs showing the histology of fetal liver from the E13.5 embryos. Details of the liver sections of syno$^{+/+}$, syno$^{+/-}$, and syno$^{-/-}$ E13.5 are shown (x 400). Cell density was found to be low in the syno$^{-/-}$ embryo.

Please amend the paragraph beginning at page 22, lines 30-35 as follows:

To analyze the status of primary hematopoiesis, peripheral blood samples obtained from syno$^{-/-}$ embryos at E10.5 were first stained and then compared with the erythroblasts formation in syno$^{+/+}$ and syno$^{+/-}$ littermates (3.0±0.66x 10^5 cells) (Fig. 5). The syno$^{-/-}$ embryos (8.3±0.46x 10^5 cells) showed diminished erythroblasts formation compared with the syno$^{+/+}$ and syno$^{+/-}$ embryos. Moreover, apoptosis was observed in syno$^{-/-}$ erythrocytes, but not in syno$^{+/+}$ erythroblasts (data not shown).

Please amend the paragraph beginning at page 23, lines 13-22 as follows:

In the above observation, apoptosis of erythroblasts observed during primary hematopoiesis in the syno$^{-/-}$ embryos was hardly observed at E12.5. To examine the reason behind this observation, the ~~liver was~~ cytocentrifuge preparation of fetal liver were stained by Giemsa. The Giemsa staining (lower row of Fig. 6) showed that images of erythroblast phagocytosis by macrophages, i.e., hemophagocytosis (lower row of Fig. 6, indicated by arrows), were hardly observed in syno$^{+/+}$ and syno$^{+/-}$, whereas in syno$^{-/-}$, such images were significantly increased and were found in 22 out of 100 macrophages (Table 3). Therefore, erythroblasts that have undergone apoptosis similar to that observed at E10.5 were engulfed and eliminated by macrophages in the liver, which is the site of hematopoiesis, and as result, it is projected that erythroblasts that have undergone apoptosis are not found in the peripheral blood derived from E12.5.

Please amend Table 2 at page 23 as follows:

[Table 2]

Erythroblast morphology	syno +/+	syno +/-	syno -/-
Nuclear disruption	0.0%	0.6%	2.0%
Binucleation	0.1%	1.2%	2.1%
Howell-Jolly body	1.7%	3.8%	6.4%

Please amend Table 3 at page 23 as follows:

[Table 3]

Mϕ phagocytosis image	syno +/+	syno +/-	syno -/-
(cells/ 100Mϕ)	1	7	22